The role of the cytoskeleton in regulating mitochondrial function in cardiomyopathic states

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Cardiomyopathy is associated with alterations in calcium homeostasis and disrupted cytoskeletal architecture. We have previously demonstrated that the L-type calcium channel (LTCC) can regulate mitochondrial function through alterations in intracellular calcium and via the transmission of movement of cytoskeletal proteins associated with channel activation and inactivation. To determine whether the cytoskeleton is integral to this process, we investigated the role of the LTCC in regulating mitochondrial function in 30-50 wk old mice over expressing the human disease causing mutations Gly203Ser (cTnI-G203S) or Arg403Gln ($\alpha MHC^{403/+}$), and exhibit characteristic features of Familial Hypertrophic Cardiomyopathy. LTCC current density recorded in cTnI-G203S and $\alpha MHC^{403/+}$ myocytes was similar to *wt* myocytes. However the inactivation rate of the current was significantly faster in cTnI-G203S and $\alpha MHC^{403/+}$ myocytes (cTnI-G203S: 22.9 ± 1.2 ms, n = 11 and MHC403: 20.2 ± 1.4 ms, n = 6 vs *wt*: 30.6 ± 1.4 ms, n = 7; P < 0 .05). We examined the effect of activation of LTCC on mitochondrial membrane potential assessed as changes in JC-1 fluorescence. Application of BayK(-) caused a 1 8.8 ± 2.0% increase in mitochondrial membrane potential in *wt* myocytes (n = 10). In contrast, BayK(-) caused a significantly greater increase in mitochondrial membrane potential in cTnI-G203S and $\alpha MHC^{403/+}$ myocytes (cTnI-G203S: 29.2 ± 1.9 ms, n = 15; MHC403: 27.8 ± 3.2 ms, n = 9; P < 0.05). Consistent with this finding, application of BayK(-) caused a significantly greater increase in flavoprotein oxidation (an indirect measure of mitochondrial oxygen consumption) in cTnI-G203S and $\alpha MHC^{403/+}$ myocytes (cTnI-G203S: 24.4 ± 6.5%, n = 8; MHC403: 24.6 ± 3.8%, n = 7) compared with *wt* myocytes (10.0 ± 1.6% increase, n = 9; P < 0.05) . These data indicate that L-type calcium channel current kinetics are altered in hearts of cTnI-G203S and $\alpha MHC^{403/+}$ mice. The mice appear to display a hypermetabolic mitochondrial state in response to LTCC activation. These findings are in direct contrast to responses recorded in myocytes from *mdx* hearts, the murine model of Duchenne Muscular Dystrophy that demonstrate poor metabolic activity and dilated cardiomyopathy. We conclude that disruption of the cytoskeletal architecture contributes to altered regulation of mitochondria by the L-type calcium channel in the cardiomyopathic heart.